92. The method of treatment according to claim 91 wherein the following effective amounts are administered (calculated on the therapeutically active compound): paclitaxel and albumin 70-280 mg per treatment; propofol and albumin 6-10 mg per kG per hour; camptothecin and albumin, gemfibrozil and albumin, cyclosporin A and albumin 3-5 mg per kG per day; amphotericin B and albumin up to 1.5 mg per kG per day;

REMARKS

The Office Action mailed March 16, 1999 has been carefully considered. Reconsideration and withdrawal of the objections to and rejections of this application are respectfully requested in view of this amendment and the following remarks.

Claims 1-23 and 38-41 have been canceled herewith. Claims 24-29 are withdrawn from consideration. Applicants submit herewith on excerpt from the textbook Immunology: E. S. Golub, Immunology: A Synthesis, 104-106 (1987) referenced herein.

New claims 42-71 have been added by amendment herewith. Support for these claims may be found throughout the specification.

Referring now to the Office Action, the Examiner notes that this present application is a continuation of PCT Application No. PCT/HU98/00086, filed on 09/18/97 and states that a reference to the prior application must be inserted as the first

sentence of the specification of this application if Applicant intends to rely on the filing date of the prior application under 3 5 U. S. C. 120. See 3 7 CFR 1. 78(a).

Applicants direct the Examiner's attention to page 2 of the Patent Application Transmittal Papers filed with the present application where the Applicants request that specification be amended by inserting before the first line thereof the following: --This application is a Continuation of PCT International Application No. PCT/HU98/00086 filed on September 17, 1998, which designated the United States and on which priority is claimed under 35 U.S.C. § 120, the entire contents of which are hereby incorporated by reference.-- In view of the foregoing, withdrawal of this objection is believed to be warranted.

The Examiner noted that there are 9 figures in this Application, and that there is no section titled "Brief Description of the Figures" describing the figures. Applicants respectfully assert that the Figures of the present application are properly described throughout the specification, for example Figure 2 is described on page 21, lines 15-19; Figure 3 is described on page 22, line 40 to page 23, line 2; Figure 4 is described on page 24, lines 13-14; Figure 5 is described on 25, lines 11-15; Figure 6 is described on page 17, lines 29-32; Figures 7, 8 and 9 are described on page 19, lines 9-16.

The term "Figure 1" appears in the specification as filed as the caption of the figure. The specification however refers to "the general formula I". It is clear from the context the term "the general formula I" refers to "Figure I" (it is the only general formula in the specification). For the purpose of clarity we have amended the

specification at page 5, line 13; page 5, line 4; and page 12, line 15, replacing "the general formula" with --Figure-- is for the sake of clarity and does not add any new matter. Support for this clarification may be found in the specification. For example, page 5, lines 11-13 discloses a formulation containing a taxonoid of the general formula I. Since a taxonoid of formula I is disclosed and Figure I is the structure of a taxonoid, it would be evident to one of ordinary skill in the art that the taxonoid of formula I is the taxonoid of Figure I. Thus, the insertion of --Figure-- is for clarity only and does not add any new matter.

The present amendments to the specification comply with M.P.E.P. §2163.07 (7th edition, 1998) which states:

> An amendment to correct an obvious error does not constitute new matter where one skilled in the art would not only recognize the existence of error in the specification, but also the appropriate correction. *In re Oda*, 443 F.2d 1200, 170 USPQ 260 (CCPA 1971).

Accordingly, these amendments do not constitute new matter.

Referring now to the claims, claims 1-23 and 30-41 are rejected under 35 U.S.C. 112, first paragraph. The Examiner states that the specification, while being enabling for a water soluble composition comprising the compounds recited in claim 8, said compounds attached to human serum albumin in a non-covalent fashion, is nonenabling for "all" possible water -soluble compositions comprising the substances recited in claim 7, linked non-covalently to "all" possible animal plasma proteins. The Examiner argues

that the specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

The Examiner takes the position that the instant specification is enabling for composition comprising a drug having low water solubility, attached to albumin, said composition retaining biological activity both in vitro and in vivo and thus the instant specification is only enabling for water-soluble compositions comprising, drugs having low water solubility and having binding affinity for albumin, attached to albumin. The Examiner further states that "absent further guidance from the specification, it would require undue experimentation, to delineate if the all the agents recited in claim 7 have binding affinities for all the proteins recited in claim 6, (glycoproteins, interferons, or interleukins), conduct the necessary experiments to show that indeed these agents do bind to said proteins, test if the composition comprising drug-protein, retain biological activity and become water soluble". The Examiner concedes that the instant specification is enabling for a composition comprising drugs bound to immunoglobulin, the specification discloses compositions attached to immunoglobulin, (see example II.4 on page 17), and shows that said compositions are active and water soluble. The Examiner further states that the instant specification, does not disclose compositions comprising drugs attached to interferon, interleukins or glycoproteins.

Applicants respectfully assert that the specification does enable a person skilled in the art to make and use the invention commensurate in scope with the present pending

claims. New pending claims 42-92 are directed to pharmaceutical compositions comprising a water soluble solid or a true aqueous solution thereof comprising: (a) a therapeutically active compound having a aqueous solubility of less than 1.10^{-4}M and having a substantial binding affinity for plasma proteins; and (b) a plasma protein in controlled aggregation state, wherein said therapeutically active compound and said plasma protein are non-covalently bound; wherein said pharmaceutical composition does not comprise an organic solvent.

The presently claimed compositions provided a new and delivery system for the administration of an active compounds with poor water solubility and provide a new means to administer effective doses of water-insoluble active compounds without introducing toxic elements (such as organic solvents) (Specification, page 3, lines 13-25) Specifically, the present invention provides for pharmaceutical compositions, the pharmaceutically active compounds having low aqueous solubility and a substantial binding affinity to plasma proteins.

Applicants respectfully disagree that one of ordinary skill in the art would require undue experimentation to determine which therapeutically active compounds are suitable for the claimed compositions. Methods for determining binding affinity are well known in the art. *See* Immunology: E. S. Golub, Immunology: A Synthesis, 104-106 (1987), attached hereto. One of ordinary skill could apply them to compounds of interest in a simple, straightforward, easy to perform and relatively inexpensive assay. Example 1 of the present application details methods to determine the binding affinity of

a particular therapeutically active compound to a particular plasma protein (Specification, pages 14 and 15). The method at Example I enables one to determine the binding of any therapeutically active compound with a plasma protein. Such experimentation is not "undue". Plasma proteins suitable for the present invention are disclosed in the specification (Specification, page 4, lines 12-22). Example II show the application of such assays to serum proteins other than albumin, including glycoproteins, interferons and interleukins (Specification, page 19, lines 25-27). The aqueous solubility of therapeutically active compounds are available in the literature (e.g. Beilstein, The Merck Index) and easily measured. Thus, the present specification enables the selection of therapeutically active compounds having an aqueous solubility of less than 1.10^{-4}M and a substantial binding affinity to plasma proteins as claimed in claims 42-92 without undue experimentation.

Claims 1-23 and 30-41 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. The Examiner also states that the term "substantial" in claim 1 is vague.

Although applicants have canceled claims 1-23 and 30-41 the term "substantial binding affinity" remains in all the pending claims. Applicants respectfully contend that newly submitted claims 43-92 particularly point out and distinctly claim the present invention in language conforming to U.S.P.T.O. guidelines. Applicants respectfully assert that the term "substantial binding affinity" is not vague. "Substantial binding

affinity" is defined ate page 3, lines 34-37 of the specification as ". . . that greater than 90% of the active substance is bound to the protein in aqueous medium in spontaneous equilibrium at room temperature". Accordingly applicants request that the 112, second paragraph rejection not be applied to the pending claims.

Claims 1-11, 21-23, 30-31, 36, 38, 40-41 are rejected under 35 U.S.C. 102 (b), as being anticipated by Satoh *et al.*, EP 0326 618 ("Satoh"). Claims 1, 12-20, 32, 34-35, 37, 39-41 are also rejected under 35 U.S.C. 103(a) as being unpatentable over Satoh.

For this rejection, the Examiner interpreted claim 1 as reciting: a water soluble pharmaceutical formulation in liquid or solid from, said formulation containing a therapeutically active compound (having low aqueous solubility, and having binding affinity to plasma proteins), bound to plasma protein.

The Examiner states that Satoh teaches a pharmaceutical composition comprising a drug having very low solubility in water, and having a protein binding property, said drug attached by hydrophobic bonding to albumin, and is in a solid or liquid form, wherein said albumin is human derived albumin, wherein the solvent is removed by distillation, said pharmaceutical composition, also contains nontoxic pharmaceutically acceptable carriers, preservatives and stabilizers. The Examiner further states that Satoh also discloses a method of treatment comprising administering said pharmaceutical composition as medicine and that the pharmaceutical composition taught by Satoh comprises 1-200 part by weight of the and 100 parts by weight of the albumin. The Examiner takes the position that Satoh teaches a pharmaceutical composition comprising

the specific drugs recited in claims 8, 11, 30 and 36 attached to albumin and therefore the Satoh reference anticipates claims 1-11, 21-23, 30-31, 36, 38, 40-41, because it teaches a pharmaceutical composition with all the limitations recited in said claims.

The Examiner concedes that Satoh does not disclose a pharmaceutical composition comprising the specific drugs recited in claims 12-20, 32-35 and 37, or a method of parenterally administrating said composition. The Examiner takes the position however that these claims would have been obvious in view of Satoh.

In the interest of facilitating prosecution, applicants have canceled claims 1-23 and 30-41. New independent claim 42 is directed to compositions suitable for parenteral administration comprising a water soluble solid or a true aqueous solution thereof comprising: (a) a therapeutically active compound having a aqueous solubility of less than 1.10^{-4}M and having a substantial binding affinity to plasma proteins; and (b) a plasma protein in controlled aggregation state, wherein said therapeutically active compound and said plasma protein are a non-covalently bound, wherein said pharmaceutical composition does not comprise an organic solvent. Applicants respectfully assert that the newly added claims are neither anticipated nor obvious in view of Satoh. Satoh discloses aqueous **suspensions** not true clear aqueous **solutions** suitable for parenteral administration. Satoh teaches only that his formulations are suitable for oral administration (Satoh, page 7, last line to page 8, 10). Satoh's compositions, when administered in liquid form, are suspensions (Satoh, page 12, lines 6-9). Satoh's formulations are not even completely dissolved during the manufacturing

process (Satoh, page 12, lines 6-9). Parenteral administration, for obvious reasons, requires a formulation of completely dissolved components. Satoh makes no suggestion as to making a formulation comprising completely dissolved components. Further, Satoh makes no suggestion as to making a formulation suitable for parenteral administration, and thereby makes no suggestion as to making a formulation comprising a therapeutically active compound *having a aqueous solubility of less than $1.10^{-4}M$ and a substantial binding affinity for plasma proteins*; suitable for parenteral administration. Satoh does not anticipate the pending claims.

The present invention in the newly submitted claims provides compositions comprising a therapeutically active compound *having a aqueous solubility of less than $1.10^{-4}M$ and a substantial binding affinity for plasma proteins*; and (b) a plasma protein in controlled aggregation state, wherein said therapeutically active compound and said plasma protein are non-covalently bound, wherein said pharmaceutical composition does not comprise an organic solvent.

Satoh does not teach or suggest the importance of selecting therapeutically active compounds having substantial binding affinity for plasma proteins. Satoh does not teach or suggest how to make the aqueous solutions of therapeutically active compounds having solubilities of less than 10^{-4} M. The claimed invention is not obvious over Satoh.

Claims 1-9 are rejected under 35 U.S.C. 102(b), as being anticipated by Mitsuharu, Inaba, JP 58-216126 (" JP '126"). The Examiner states that JP '126 teaches a

composition comprising water-insoluble drug and human serum albumin, said drug becoming soluble after it is dissolved in a solution comprising human serum albumin and water. The Examiner takes the position that claims 1-9 of the present application are drawn to a pharmaceutical composition comprising a water insoluble drug and albumin and therefore JP '126 anticipates the instant claims 1-9 in the absence of any evidence to the contrary.

Applicants respectfully assert that JP '126 does not anticipate the newly submitted pending claims 42-92. The abstract of, JP '126 (referred to by the Examiner and attached to the PTO Form 1449) discloses in full:

> Purpose: An auxiliary for dissolution handleable by simple operations, making a water-insoluble or water slightly soluble drug dissolve in water easily, comprising human serum albumin.
> Constitution: A water-insoluble or water slightly soluble drug is added to a diluted aqueous solution of human serum albumin, so that the drug is solubilized. A long-chain type carboxylic acid having an aromatic functional group with large steric hindrance, such as (5Z,15xsi)- 9,11 -methano-10,10-dimethyl-13-aza-15-hydroxy-16-phenyl-17,18,19,20-tetranor-11a-carbathrombo-5-enoic acid, a thromboxane A2 antagonist, not long-chain type carboxylic acid, such as (E)-3-[4- (pyridine-3-ylmethyl)]phenylmethacrylic acid, a thromboxane A2 biosynthesis inhibitor, an unfatty acid (e.g., steroid, barbiturate), etc. may be cited as the drug, and the auxiliary is effective for a drug having <=0.1mg/ml water solubility.

JP '126 abstract discloses that human serum albumin serves as a solubilizer for certain special compounds: carboxylic acids containing highly hindered aromatic functional groups, certain steroids, barbiturates. No explanation is given as to what common properties the members of the group share. The method of the '126 abstract is

not workable for the highly insoluble compounds of the present invention having the low aqueous solubilities indication therein so as to achieve solutions containing effective amounts of the drugs. No clear solutions can be so obtained.

Applicants respectfully submit that the abstract of JP '126 does not anticipate the pending claims. The abstract states that the "auxiliary" for dissolution is effective for a drug having <= 0.1 mg/ml water solubility. However, the pending claims are limited to drugs having aqueous solubility of less than $1x10^{-4}$M. Assuming that the compounds of JP '126 have a molecular weight in the range of 100-300, 0.1 mg/mL corresponds to 10^{-3} to 3×10^{-4} -- outside the scope of the pending claims. Thus JP '126 does not anticipate.

Furthermore, JP '126 is not enabled for the pending claimed invention. JP '126 abstract discloses a process of adding the solid compound to a diluted aqueous solution of human serum albumin. Thus for at least these reasons, this non-enabling abstract cannot render the pending claims obvious.

Claims 1-10 are rejected under 35 U.S.C. 102 (e), as being anticipated by U.S. Patent No. 5,916,596 to Desai *et al* ("Desai"). The Examiner states that Desai teaches a pharmacologically active agent in combination with albumin. The Examiner further states that Desai teaches a pharmaceutical composition comprising taxol or paclitaxel and albumin, said composition having low toxicity, and said composition administered by intramuscular, or intravenous route of administration. The Examiner takes the position that claims 1-10 of the instant application are drawn to a water-soluble composition composing an active compound and plasma protein, said composition

administered parenterally and therefore Desai anticipates the instant claims 1-10 in the absence of any evidence to the contrary.

In Desai's compositions, the pharmacologically active agent is contained in a poly-(albumin) shell where the polymer shell is cross-linked as a result of exposure to high shear conditions (Desai, col 8, lines 29-38). Applicants respectfully assert that Desai does not anticipate the newly submitted pending claims. Desai does not disclose a therapeutically active compound having an aqueous solubility of less than 1.10^{-4} M and having a substantial binding affinity for plasma proteins where the therapeutically active compound is non-covalently bound to a plasma protein. Rather, Desai discloses a therapeutically active compound contained within a polymeric shell. The therapeutically active compound is not bound to the shell; the shell is not a plasma protein (it is described as a cross-linked polymer of plasma protein monomers). Thus, Desai does not anticipate a water soluble composition comprising a therapeutically active agent having an aqueous solubility of less than 1.10^{-4} M and having a substantial binding affinity for plasma proteins wherein the therapeutically active compound is non-covalently bound to a serum protein. Therefore, Desai does not anticipate the present pending claims 42-92. Nor does Desai render the pending claims obvious. Desai teaches a different method of solubilizing water-insoluble materials by encasing them in a water soluble polymer shell. It is irrelevant to Desai's compositions whether the water insoluble compounds have a substantial binding affinity for plasma proteins. His compositions would appear to work with any water insoluble therapeutic compound capable of being encapsulated,

regardless of their ability to bind to plasma proteins. Therefore Desai does not render applicant's invention obvious.

In view of the foregoing, Applicants submit that all of the pending claims of the subject application are now in condition for allowance, and issuance of a Notice of Allowance is respectfully requested. The Examiner is invited to telephone the undersigned attorney at (212) 908-6305 if there are any questions concerning this amendment.

Respectfully submitted,

Date: Sept. 18, 2000

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IMMUNOLOGY
A SYNTHESIS

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THE COVER

Computer graphic view of insulin showing that antigenic determinants cluster at highly flexible regions. The alpha carbon backbone is shown with purple lines. The molecular surface is indicated with dots, color-coded from most mobile to most rigid in the order red, yellow, green, cyan, blue. Residues forming antigenic determinants are labeled in pink (contiguous) and yellow (discontiguous) and can be seen to correspond with the more mobile regions (see pages 28 and 29). [Image created by John A. Tainer and Elizabeth D. Getzoff, Research Institute of Scripps Clinic]

PART-OPENING ELECTRON MICROGRAPHS

Part One, p. 15: Antibody–hapten complex (purified rabbit anti–2,4-dinitrophenyl antibody and a bivalent hapten). [From R. C. Valentine and N. M. Green (1967), *J. Mol. Biol.* 27, 615]

Part Two, p. 155: A resting lymphocyte, probably a T cell, ×21,840. [Courtesy of D. Zucker-Franklin, New York University Medical Center]

Part Three, p. 437: Immune complexes, seen as electron-dense, hump-shaped deposits in the upper third of the photo, along a capillary wall in a glomerulus following streptococcal glomerulonephritis (×17,250). [Courtesy of M. N. Yum, Indiana University Medical Center]

IMMUNOLOGY: A SYNTHESIS

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CHAPTER SEVEN

The interaction of an antibody with an antigen is no different in principle from any other bimolecular reaction between a ligand and a molecule that specifically binds that ligand. The reaction between enzyme and substrate is a bimolecular reaction and follows the same rules of physical chemistry as do antigen–antibody reactions. The difference between the two is that the substrate is changed in an enzyme–substrate reaction, but not in an antigen–antibody reaction. A reaction in which one of the reactants is altered will be a one-way, or nonreversible, reaction. Antigen–antibody reactions are reversible because the interaction does not result in permanent change to either of the reactants.

The Antigen–Antibody Complex

The reaction of antigen with antibody results in the formation of an ANTIGEN–ANTIBODY COMPLEX (AGAB):

$$Ag + Ab \rightleftharpoons AgAb \quad (1)$$

AgAb is formed by *noncovalent* interactions such as hydrogen bonding, polar or hydrophobic bonding, ionic or coulombic interaction, and van der Waals forces. Because the reaction is noncovalent and neither reactant is altered, the reaction is in theory a reversible one. All of the methods used to quantify the antigen–antibody reaction depend upon the ability to measure AgAb.

We will see in the next chapter that the form taken by AgAb depends on the nature of the antigen. Soluble antigens such as proteins form complexes with antigen and become insoluble precipitates. Particulate antigens, such as cells, after reacting with antibody may form an AgAb that agglutinates. The reaction is analyzed by determining either the rate or quantity of precipitate or agglutinate.

Affinity of the Antigen–Antibody Reaction

Because the formation of AgAb can be treated as a chemical reaction between two ligands and because the reaction is reversible, the *affinity*, or strength, of the reaction can be determined. The law of mass action states that the rate of a reaction is propor-

tional to the concentration of the reactants. By applying the law of mass action to equation (1) we obtain

$$\mathrm{Ag} + \mathrm{Ab} \underset{k_d}{\overset{k_a}{\rightleftharpoons}} \mathrm{AgAb} \qquad (2)$$

$$k_a[\mathrm{Ab}][\mathrm{Ag}] = k_d[\mathrm{AgAb}]$$

where [Ag] and [Ab] are the concentrations of free antigen and antibody; [AgAb] is the concentration of bound Ag and Ab, that is, the AgAb complex; and k_a and k_d are the association and dissociation constants.

From equation (2) we can arrive at the *equilibrium constant* for the reaction:

$$K = \frac{k_a}{k_d} = \frac{[\mathrm{AgAb}]}{[\mathrm{Ag}][\mathrm{Ab}]} \qquad (3)$$

AFFINITY is the sum of the noncovalent attractive and repulsive forces stabilizing the complex and is therefore the same as the EQUILIBRIUM CONSTANT K (which is expressed in liters/mole).

These reactions hold only for homogeneous binding sites (antibodies) and ligands (haptens). We know from the preceding chapters that antibodies are extremely heterogeneous; therefore these equations are only an approximation of the actual conditions. For monoclonal antibodies, however, these equations come very close to representing the actual interactions.

Determining Affinity

To determine the affinity of the reaction, we obviously need a means of determining the concentration of free antigen and bound hapten. This is most conveniently accomplished by EQUILIBRIUM DIALYSIS. This method is diagrammed in Figure 1. Antibody and hapten are separated by a semipermeable membrane. The hapten, with a suitable label (such as a radioisotope) is placed on one side of the membrane and the antibody is placed on the other. The pore size of the membrane is such that the hapten freely passes through; but the antibody, being of higher molecular weight, does not. Samples are then taken from each side at various times to determine the amount of hapten on each side.

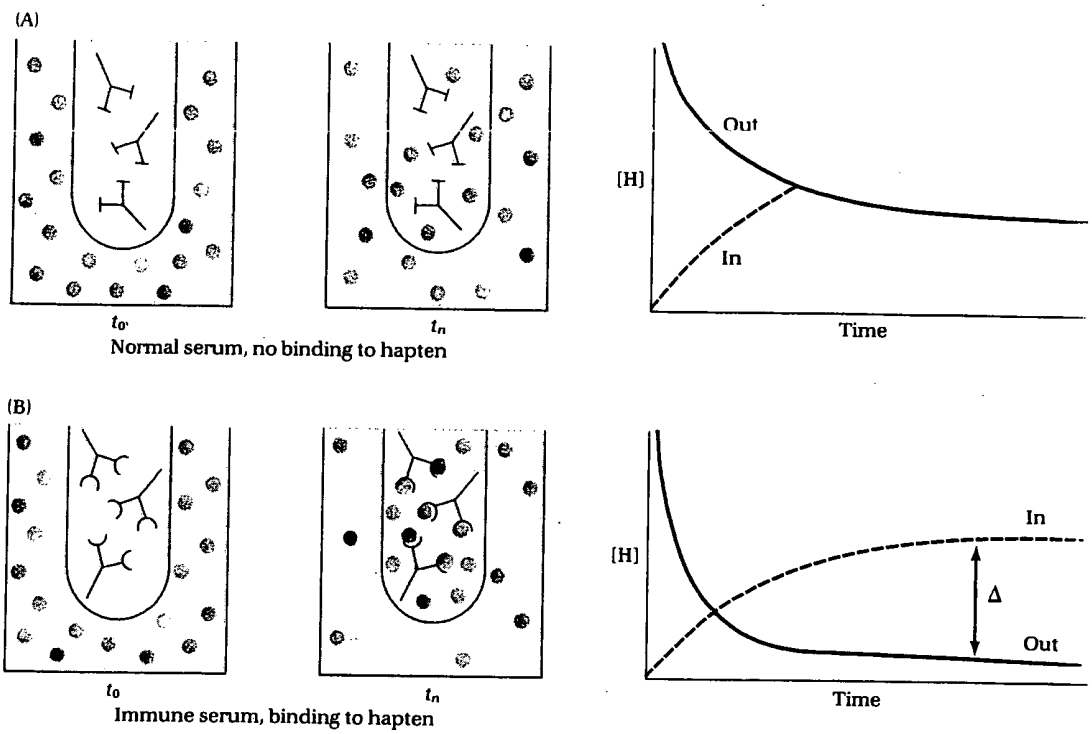

FIGURE 1 EQUILIBRIUM DIALYSIS

Anti-hapten antibody and labeled hapten are separated by a dialysis membrane. With time the hapten molecules diffuse across the membrane and reach equilibrium. (A) In normal serum no binding of the hapten occurs when it crosses the membrane. At equilibrium equal amounts of hapten are present on both sides of the membrane. (B) In immune serum the hapten is bound by the anti-hapten antibody, reducing the concentration of free hapten [H] inside. When the unbound hapten reaches equilibrium, there is a difference Δ between [H] in and [H] out. The difference Δ is the amount of hapten bound.

In Figure 1A the antibody is not directed to the hapten, so no AgAb forms and all of the hapten is unbound. This condition serves as a control because it tells the rate at which the concentration of the hapten reaches equilibrium on both sides of the membrane. In Figure 1B the antibody reacts with the hapten, so that some of the hapten moving into the antibody compartment